B2 CONT substantially identical to MYB polypeptides exemplified here (*e.g.*, SEQ ID NOS:2, 4, 6 or 8). The polynucleotide can be, for example, SEQ ID NOS:1, 3, 5 or 7.--

Please replace the paragraph beginning at page 4, line 14, with the following:

--The term "MYB polynucleotide" refers to a polynucleotides encoding a member of a class of transcription factors referred to here as "MYB polypeptides". MYB polypeptides are characterized by the presence of an amino-terminal DNA-binding domain, or DBD, consisting of two or three helix-turn-helix motifs of 51-52 amino acids (R1, R2 and R3) that are highly conserved across phyla. MYB polypeptides may also comprise a transactivation domain. Exemplary MYB polypeptides are disclosed in SEQ ID NO:1 (GhMYB 1 GenBank Accession No. L04497) and SEQ ID NO:3 (GhMYB 6 GenBank Accession No. AF034134). Other useful sequences include

B3 sequences at GenBank Nos. AF034130 (GhMYB 2), AF034131 (GhMYB 3), AF034132 (GhMYB 4), and AF034133 (GhMYB 5). In addition, two other MYB nucleotide sequences are provided (GhMYB 7 and 8 (SEQ ID NOS:5 and 7). One of skill in the art will recognize that in light of the present disclosure, various modifications (*e.g.*, substitutions, additions, and deletions) can be made to the MYB polypeptide sequences without substantially affecting their function. For example, the MYB polypeptides may contain functional domains from other porteins (*e.g.* related MYB polypeptides). These variations are within the scope of the term "MYB polypeptide". For example a MYB polypeptide includes the sequences exemplified here as well as polypeptides that are at least about 60%, usually at least about 70%, more usually at least about 80%, and often at least about 90% identical to the exemplified sequences. Also included are variant nucleic acid sequences that encode the same polypeptide as the exemplified sequences, *i.e.* sequences comprising degenerate sequences.--

Please replace the paragraph beginning at page 11, line 16, with the following:

--Alternatively, the nucleic acids of interest can be amplified from nucleic acid samples using amplification techniques. For instance, polymerase chain reaction (PCR) technology

B4 can be used to amplify the sequences of genes directly from genomic DNA, from cDNA, from genomic libraries or cDNA libraries. PCR and other *in vitro* amplification methods may also be

useful, for example, to clone nucleic acid sequences that code for proteins to be expressed, to make nucleic acids to use as probes for detecting the presence of the desired mRNA in samples, for nucleic acid sequencing, or for other purposes. For a general overview of PCR, *see PCR Protocols: A Guide to Methods and Applications*. (Innis, M, Gelfand, D., Sninsky, J. and White, T., eds.), *Academic Press*, San Diego (1990). Appropriate primers and probes for identifying sequences from plant tissues are generated from comparisons of the sequences provided herein (*e.g.* SEQ ID NOS:1 and 3).--

B4 cont

Please replace the paragraph beginning at page 12, line 2, with the following:

--The isolated nucleic acid sequences prepared as described herein can be used in a number of techniques. For example, the isolated nucleic acids can be introduced into plants to enhance endogenous MYB gene expression and thereby increase expression of the genes whose expression is controlled by MYB polypeptides. A particularly useful gene for this purpose are the MYB genes shown in SEQ ID NOS:1 and 3.--

B5

Please replace the paragraph beginning at page 21, line 9, with the following:

--An *EcoRI/XhoI* restriction fragment containing the *Gl1* gene (Oppenheimer, D.G. *et al., Cell* **67**:483-493 (1991)) was initially used to screen an unamplified gt10-3 dpa cotton ovule cDNA library (Wilkins, T.A. *et al., Plant Physiol* **102**:679-680 (1993)). Because only one *MYB* clone *(GhMYB1)* was recovered, a heterogeneous pool of homologous DNA probes spanning the conserved MYB DNA-binding domain (DBD) was generated by PCR for a second round of library screening. To amplify 1 59-bp of the DBD, two degenerate 'universal' *MYB* primers, COT20 COT2O (5'-GGNAARAGYTGYMGITTRAG-3'; SEQ ID NO:9 5'-GGNAARAGYTGYMGITTRAG-3") and COT21 (3'-GGNCCKKCTTGTCTRTTRS-5'; SEQ ID NO:10) were designed against the highly conserved stretches coding for peptides GKSCRL (SEQ ID NO:11) and PGRTDN (SEQ ID NO:12), respectively. Using 25 µl of recombinant phage (3.4×10^7 pfu/µl) from the same unamplified library as the template, a 125 µl reaction was set-up containing 1x reaction buffer, 0.4 mM dNTPs, 0.5 µM of each COT primer, and 0.04 units of Promega *Taq* DNA polymerase/µl, with the final concentration of 1.5 mM $MgCl_2$ provided by the phage storage

B6

buffer. PCR was performed in a thermal cycler (Ericomp) as follows: 30 sec at 94°C (1 cycle); 2 min at 92°C, 2 min at 48°C, 2 min at 72°C (25 cycles); and 10 min at 72°C (1 cycle). The 159-bp amplicon was cloned into the pT7Blue TA-cloning vector and transformed into *E. coli* NovaBlue competent cells (Novagen). Nucleotide sequencing (Sanger, F. *et al., Proc Natl Acad Sci USA* **74**:5463-5467 (1977)) of 24 independent transformants identified a total of six different *MYB* DBDs. Equal amounts of the 159-bp amplicon from each of the six PCR clones, released by *EcoRI* digestion, were combined in a heterogeneous pool of DBD sequences for use as a homologous hybridization probe. The pooled DNA probes were radiolabeled with [^{32}P]-dATP by random-primer labeling (Feinberg, A.P. *et al., Anal Biochem* **132**:6-13 (1983)). Two sets of plaque lifts containing 3-4 x 10^5 recombinant phage from an amplified gt10-3 dpa cotton ovule gtl0 cDNA library (Wilkins, T.A. *et al., Plant Physiol* **102**:679-680 (1993)) were prepared using Hybond-N nylon membranes (Amersham). Both sets of plaque lifts were hybridized overnight at 42°C in 50% formamide buffer according to the membrane manufacturer's instructions. Lifts hybridized to the Arabidopsis *AtMYBG/1* probe were washed in 2x SSC, 0.1% SDS at 60°C for 30 min (moderate stringency), whereas lifts hybridized to the heterogeneous pool of cotton DBDs were washed in 0.2x SSC, 0.1% SDS at 60°C (high stringency). DNA prepared from 15 purified recombinant-phage plaques (Sambrook, J. *et al., Molecular cloning: a laboratory manual. Cold Spring Harbor Laboratory Press Cold Spring Harbor NY 2nd Edn.* (1989)) were *Eco*RI digested and electrophoresed in low melting point agarose. They were subcloned into pUC118 using the excised gel slices directly in the ligation reaction (Struhl, K. *Biotechniques* **3**:452-453 (1985)) and transformed into *E. coli* MV1190. The complete nucleotide sequence (Sanger, F. *et al., Proc Natl Acad Sci USA* **74**:5463-5467 (1977)) of both strands was determined for each of the 15 *GhMYB* isolates (designated A through O), using Sequenase 2.0 (US Biochemical) or the Klenow fragment of DNA Polymerase I (GibcoBRL) in overlapping nested deletions of single-stranded DNA as templates (Dale, R. *et al., Methods Enzymol* **155**:205-231 (1987)). To comply with standard nomenclature for *R2R3-MYB* genes, the six unique cotton *MYB* clones *A, D, G, J, N* and O were renamed and numbered sequentially as *GhMYB1* through *GhMYB6,* respectively.--

B6 cont

Please replace the paragraph beginning at page 22, line 28, with the following:

--Genomic DNA was extracted from young expanding leaves of *G. hirsutum* L. as described previously (Wilkins, T.A. *et al., Theor Appl Genet* **89**:514-524 (1994)). CsCI-purified DNA (20 µg) was completely digested with *EcoRI, Hind*III or *Hind*II, fractionated in 1% agarose gels at 1.8 V/cm for 12-15 h, and transferred to Zetabind nylon membrane *(Cuno,* Meriden, CT) by capillary blotting in 10x SSC transfer buffer. Afterwards, the DNA was affixed to the membrane matrix by UV cross-linking. Membrane hybridization was performed at 65°C in 5x SSPE, 7% SDS, and 250 µg/ml of sheared, denatured salmon sperm DNA. Gene-specific hybridization probes were generated by PCR, using oligonucleotide primer-pairs designed against unique nucleotide sequences of the six cotton *MYB* cDNAs *(GhMYBI-6)*. The primers (Operon Technologies), with respective sizes of amplified fragments were: COT105 (5'-AAGCAGAGGAATTGATCCAC-3'; SEQ ID NO:13) x COT106 (3'-CTGGGAACCTAAGTATCCCA-5'; SEQ ID NO:14), 538 bp; COT107 (5'-CCTCGGAACAAATTGTGCC-3'; SEQ ID NO:15) x COT108 (3'-GCCTTCCAACGAAACCAAACC-5'; SEQ ID NO:16), 153 bp; COT109 (5'-CAGAAGGAGAAACACAGAGG-3'; SEQ ID NO:17) x COT110 (3'-GGCTGTATCACTTGACATCG-5'; SEQ ID NO:18), 412 bp; COT111 (5'-CCATTAACTCAAAGCATGCC-3'; SEQ ID NO:19) x COT112 (3'-CGAGGAGGAACAAGGAGGAC-5'; SEQ ID NO:20), 861 bp; COT113 (5'-AGTCCAGAAGCAGGCCAAGC-3'; SEQ ID NO:21) x COT114 (3'-GGTGTACTTAAGCATTAGCA-5'; SEQ ID NO:22), 545 bp; and COT 115 (5'-CACCGCCCACTGGTATATCC-3'; SEQ ID NO:23) x COT 116 (3'-CCGTTGTACGTGCGGTAACA-5'; SEQ ID NO:24), 243 bp. The concentrations of the PCR components in a 25 µl reaction were 1x synthesis buffer, 1.5 mM $MgCl_2$, 0.2 µM of each primer, 0.2 mM dNTPs, and 0.04 units of *Taq* polymerase/µl. The temperature cycling conditions included the same denaturing and extension steps as stated previously, but 30 cycles of 1 min at 94°C, 1 min at 48°C, and 1 min at 74°C were used instead. The resulting PCR products of the expected molecular weight were purified using Promega's Magic® PCR columns, cloned into the pCR™ II T-vector (Invitrogen) and transformed into *E. coli* MV1190. These PCR fragments were radiolabeled and used as gene-specific hybridization probes. Blots were washed under high stringency conditions in 0.lx SSC, 0.1% SDS at 65°C for 30 min prior to autoradiography.--

Please replace the paragraph beginning at page 23, line 26, with the following:

--Semi-quantitative RT-PCR was performed with minor modifications (An, Y-Q. *et al., Plant Cell* **8**:5-30 (1996)) using total RNA isolated from various tissues and developing ovules by the procedure of Wan, C-Y *et al., Anal Biochem* **223**:7-12 (1994). First-strand cDNA synthesis was performed using 1.5 μg of total RNA as the template according to recommendations in the 3' RACE kit (GibcoBRL). The amount of cDNA synthesized, as determined by spectrophotometer and fluorometer measurements, indicated cDNA yields ranging from 3 to 6 x 10^2 ng per RT reaction. Semi-quantitative PCR was performed in two sequential amplification steps using ten-fold serial dilutions (10^{-1}, 10^{-2} and 10^{-3}) from the same amount of first-strand cDNA (500 pg). Attempts to include more than one primer-pair in a given reaction produced inconsistent results (data not shown), therefore expression analysis was performed independently for each *GhMYB* gene. PCR products could not be detected in dilutions greater than 10^{-3} by agarose gel electrophoresis. In the first round of amplification, PCR was performed using the universal *MYB* primer COT2O and the universal AP primer provided with the 3' RACE kit. The volume of each 10-fold RT-dilution step used as the template was 8% of the final PCR volume. In the second amplification, gene-specific primer-pairs (see above and Fig. 1b) were employed, using 8% of the previous PCR reaction as the template. The two sequential rounds of PCR amplification included lx reaction buffer, 0.2 mM dNTPs, 0.2 μM for each primer, and 0.05 units of *Taq* DNA polymerase/μl of the reaction. Temperature cycling conditions were the same as described above for gene-specific amplifications. PCR products from the second amplification were resolved in 1% agarose gels stained with ethidium bromide and visualized using a IS1000 still-video imaging system (Alpha Innotech). Recorded images were stored as TIF files. At least three replicated experiments were performed for each gene-specific primer-pair. As a reference, a 300-bp portion of the vacuolar H^+-ATPase catalytic subunit was amplified by the primer-pair [COT8 X COT9] (Wilkins *et al.,* 1994) under the same conditions to verify that the tissue-specific distribution and developmental profile produced by semi-quantitative RT-PCR was consistent with the expression pattern produced by northern blot analysis or ribonuclease protection assays (Smart, L.B. et al, *Plant Physiol* **116**:1539-1549 (1998)).--

Please replace the paragraph beginning at page 24, line 26, with the following:

--Fifteen cDNA clones encoding cotton *MYB*-domain *(GhMYB)* genes were isolated from ovules at -3 dpa, the stage at which trichome primordia are fully potentiated to develop (Wilkins, T.A. *et al., In Basra AS (ed) Cotton Fibers. Food Products Press New York* (1999)). Complete sequence analysis of the 15 ovule cDNA clones revealed the presence of six distinct *MYB*-domain genes, designated as *GhMYB1* through *GhMYB6*, based on both the nucleotide variation within the DBD and the presence of unique C-terminal domains. Among the clones characterized, three appeared to be closely related members of *GhMYB2, -3* and -6, while the remainder of the 15 clones represented additional full-length or truncated versions of the six GhMYBs. Not surprisingly, the region spanning the amino-terminal DBD was very highly conserved among all six GhMYBs, with amino acid identities/similarities ranging from a low of 54.8/16.4% (GhMYB5 vs. GhMYB6) to a high of 84.6/11.5% (GhMYB1 vs. GhMYB6). Structural similarities among the cotton and other plant MYBs includes a typical R2/R3 repeat, the tryptophan hydrophobic core and conserved DNA base-contacting residues that function in recognition specificity (Martin, C. *et al., Trends in Genet* **13**:67-73 (1997); Ogata, K. *et al., Cell* **79**:639-648 (1994)). For descriptive purposes of this work, the entire C-terminal region downstream of the DBD was designated as the transcriptional (trans-) regulatory region, or TRR. We found this designation appropriate, considering that (i) both the TAD and NRD are located in this region in animals, (ii) the relative position of the TAD within the MYB C-terminal region may vary, and (iii) the TRR region varies considerably in both the number and composition of amino acids in a MYB-specific, even within a given species (Avila, J. *et al., Plant J* **3**:553-562 (1993); Jackson, D. *et al., Plant Cell* **3**:115-125 (1991); Li, S.F. *et al., Plant J* **8**:963-972 (1995)); and (iv} the number and type f conserved motifs varies from MYB-to-MYB. Apart from a few conserved motifs, each GhMYB TRR is unique, ranging in size and amino acid composition, and in the location of putative leucine-zipper structures.--

B9

Please replace the paragraph beginning at page 25, line 18, with the following:

--Interesting structural features were identified that may have implications on regulatory aspects of MYBs. First of all with the noted exception of GhMYB2, the remaining 5 GhMYBs contain a conserved stretch of 40 amino acids with a positive net charge (basic pl) in the 5'-portion of the TRR proximal to the DBD. To our knowledge, this is the first clear description of such a basic domain outside of the DBD, and consequently led to the subdivision of the TRR into a

B10

basic 'transregulatory region 1' (TRR1), and an acidic 'transregulatory region 2' (TRR2) in these MYBs. Secondly, the presence of a conserved GIDPxxH (SEQ ID NO:25) motif was noted within the TRR1 of GhMYB1 and GhMYB6, and is located precisely 12 amino acids downstream of the last tryptophan of the R3 repeat. When present, the GIDPxxH (SEQ ID NO:25) motif is found in exactly the same position of other plant MYBs, irrespective of whether there is a TRR1 domain or not (Jackson, D. *et al., Plant Cell* **3**:115-125 (1991); Li, S.F. *et al., Plant J* **8**:963-972 (1995); Lin, Q. *et al., Plant Mol Biol* **30**:1009-1020 (1996); Marocco, A. *et al., Mol Gen Genet* **216**:183-187 (1989); Wissenbach, M. *et al., Plant J* **4**:411-422 (1993)). Analysis of the six GhMYBs and 24 additional plant MYB sequences therefore established that there was no apparent association between the presence, or absence, of this motif and TRR1. Thus, plant MYBs having a TRR1 may (GhMYB1 and -6) or may not (GhMYB3, -4 and -5) necessarily contain the GIDPxxH (SEQ ID NO:25) motif. Third GhMYB1 possesses a cysteine-rich zinc-finger motif ($CX_1CX_{10}CX_2C$ (SEQ ID NO:26), where X = any amino acid; Chopra, S. *et al., Plant Cell* **8**:149-1158 (1996)) near the carboxyl-terminus of the TRR, which confers a unique protein structure to this GhMYB by the presence of two potentially functional DNA-binding domains within a single polypeptide. Finally, the presence of small 5'-upstream open reading frames (5'-uORFs) located within the 5'-untranslated region of *GhMYB4* and *-5* are worth noting, since such uORFs have been shown to drastically interfere with the level of translation of the correct ORF of other transcription factors (Damiani, R.D. *et al., Proc Natl Acad Sci USA* **90**:8244-8248 (1993); Lohmer, S. *et al., Plant Cell* **5**:65-73 (1993)).--

B10 cont

Please replace the paragraph beginning at page 27, line 28, with the following:

--A semi-quantitative reverse transcription-polymerase chain reaction (RT-PCR) method (An, Y-Q. *et al., Plant Cell* **8**:5-30 (1996)) was employed using TRR gene-specific primer-pairs to determine the expression pattern and relative abundance of individual *GhMYB* transcripts. In control experiments, oligonucleotide primers did not amplify non-specific PCR products. As a point of reference, the transcript profile of the vacuolar H^+-ATPase H*-ATPase catalytic subunit produced by RT-PCR from the RNA source used to amplify GhMYB mRNAs was identical to published expression patterns detailing the spatial and temporal regulation of this gene (Hasenfratz, M-P *et al., Plant Physiol* **108**:1395-1404 (1995); Smart, L.B. et al, *Plant Physiol* **116**:1539-1549 (1998)). A serial dilution of each RT reaction (10^0, 10^{-1}, 10^{-2}, and 10^{-3}) was performed with a fixed

B11

B11 cont amount of first-strand cDNA to restrict the availability of template during PCR amplification. By doing so, only the more abundant messages can be detected at higher dilutions.--

Please replace the paragraph beginning at page 28, line 6, with the following:

--To evaluate the spatial pattern of *GhMYB* gene expression, semi-quantitative RT-PCR analysis was performed using total RNA from several organs and tissues. Expression of *GhJMY1*, *-2* and *-3* was detected at the 10^{-3} dilution in the all tissues and organs tested, including developing cotton fibers, although the relative abundance for these *GhMYB* transcripts was considerably lower in pollen and stigmas. In contrast, expression of *GhMYB4, -5* and *-6* varied considerably in relative abundance and was spatially regulated in a tissue-specific manner. To distinguish between the two expression patterns, the broad distribution of *GhMYB1, -2* and *-3* transcripts was termed as type I, whereas type II referred to the tissue-specific pattern of expression

B12 exhibited by *GhMYB4*, *-5,* and *-6,* which includes the absence of transcripts in stigmatic tissue. *GhMYB4* is preferentially expressed in ovules since mRNA was strongly detected at the 10^{-3} dilution in ovules + fibers, but only at $< 10^{-1}$ in isolated fibers, indicating at least a 10- to 100-fold difference in transcript abundance. Similarly. *GhMYB4* transcripts were present in roots, leaves, and petals, but in lower abundance ($<10^{-1}$) than observed in ovules. *GhMYB4* expression was not detected whatsoever in bracts, pollen, anthers or embryos (10^{0} dilution). *GhMYB5* mRNA, on the other hand, was clearly detected in bracts, and to a lesser extent in petals and anthers, but was in low abundance in roots, leaves, ovules, + fibers (10^{-2} dilution), and was barely detectable in pollen (10^{-1} dilution). *GhMYB5* transcripts in petals were of slightly lower molecular weight than expected in all experiments. Since this phenomenon has been observed to selectively occur in floral tissues using unrelated primers (Hasenfratz, M-P *et al., Plant Physiol* **108**:1395-1404 (1995)), one possible explanation for the difference in transcript size is the tissue-specific use of alternative poly(A) signals or alternative splicing, although amplification of a PCR artifact cannot be totally excluded. *GhMYB6* transcripts were strongly detected in roots, bracts, petals, anthers, ovules + fibers, and fibers, and to a lesser degree in leaves and embryos at the 10^{-3} dilution. *GhMYB6* mRNA in pollen was detected at dilutions $<10^{-2}$. Although the spatial distribution and transcript abundance of *GhMYB6* shared characteristics of both type I and type II expression patterns, and was therefore

B12 cont somewhat intermediate between the two types, *GhMYB6* was classified as type II for the time being.--

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Please replace the paragraph beginning at page 30, line 24, with the following:

--As expected, the R2R3 structure of the DNA-binding domain of the six newly identified cotton MYB genes *(GhMYB)* is highly conserved, whereas the amino acid sequence of the C-terminal domain, termed the transcriptional regulatory region (TRR), is highly variable. The TRR of GhMYB4 is considerably longer than the average plant MYB resulting in a R2R3-Myb factor of unusual molecular weight (50.8 kD). Beyond the differences in length and composition of the TRR, several other interesting structural features that may influence the target gene specificity of *GhMYBs* warrant further discussion. GhMYB1 in particular, is one distinct example in that it contains a cysteine-rich domain similar to a zinc-finger motif, $CX_1CX_{10}CX_2C$ (SEQ ID NO:26) (Chopra, S. *et al., Plant Cell* **8**:149-1158 (1996)). This MYB DBD/zinc-finger combination was first reported in a *P* allele from maize and one which exhibits a distinct tissue-specific pattern of expression (Chopra, S. *et al., Plant Cell* **8**:149-1158 (1996)). A suggested in the maize study, the presence of two potentially functional DNA-binding domains within a single polypeptide may confer a unique means for modulating gene expression. although this supposition has yet to be tested in either species.--

B13

Please replace the paragraph beginning at page 31, line 5, with the following:

--A second noteworthy feature of R2R3-MYBs identified in this study is a basic 40-amino acid region of the TRR, designated as TRR1 to distinguish this subdomain from the acidic portion (TRR2) of the C-terminus. The TRR1 which is located immediately downstream of the DBD, is found in a subset of plant MYBs or about 50% of the R2R3-MYBs examined so far (Avila, J. *et al., Plant J* **3**:553-562 (1993); Cone, K.C. *et al., Plant Cell* **5**:1795-1805 (1993); Grotewold, E. *et al., Proc Natl Acad Sci USA* **88**:4587-4591 (1991): Jackson, D. *et al., Plant Cell* **3**:115-125 (1991); Li, S.F. *et al., Plant J* **8**:963-972 (1995); Lin, Q. *et al., Plant Mol Biol* **30**:1009-1020 (1996); Noda, K-I *et al., Nature* **369**:661-664 (1994); Urao, T. *et al., Plant J* **10**:1145-1148 (1996); Wissenbach, M. *et al., Plant J* **4**:411-422 (1993)). Although the functional significance of TRR1 is not known, one might reasonably infer from the basic nature of this region that the TRR1 subdomain

B14

plays a role in modulating the interaction with DNA molecules (Mitchell, P.J. *et al., Science* **245**:371-378 (1989)). The fact that *GhMYB2* and many other plant MYBs, lack a TRR1 subdomain would suggest a degree of specialization for these particular MYB proteins in terms of DNA-binding affinity and/or sequence recognition. It was also noted that the conserved motif, GIDPxxH (SEQ ID NO:25), is present within the TRR1 domain of GhMYB1 and GhMYB6 and is located precisely 12 amino acids away from the last tryptophan of the R3 repeat in both proteins. Interestingly. while the TRR1-GIDPxxH (SEQ ID NO:25) combination identified in GhMYB1 and GhMYB6 is conserved in MIXTA (AmMYBMx). Glabrous1 (AtMYBGIl) does not contain either a TRR1 or GIDPxxH (SEQ ID NO:25) motif and is therefore structurally analogous to GhMYB2. Taken together, the combined diversity of DBD, TRR1 and TRR2 domains, plus the presence/absence of particular motifs, may endow GhMYBs with greater flexibility in the formation of functional transcription complexes (Ess, K.C. *et al., Mol Cell Biol* **15**:5707-5715 (1995); Kanei-Ishii, C. *et al., J Biol Chem* **269**:15768-15775 (1994); Oelgeschläger, M. *et al., EMBO J* **15**:2771-2780 (1996)).--

B14 cont

Please replace the paragraph beginning at page 31, line 31, with the following:

--A third interesting structural property that likely modulates expression of *GhMYB4* and *GhMYB5* at the translational level is the presence of 5'-uORFs. The effect of these small upstream open reading frames on translation is well known - resulting in decreased synthesis of the major polypeptide by interfering with re-initiation of translation at downstream start condons (Damiani, R.D. *et al., Proc Natl Acad Sci USA* **90**:8244-8248 (1993); Lohmer, S. *et al., Plant Cell* **5**:65-73 (1993)). For GhMYB5 in particular, the AUG initiation context (Dasso, M.C. *et al., Eur J Biochem* **187**:361-371 (1990); Gallie, D.R. *Annu Rev Plant Physiol Plant Mol Biol* **44**:77-105 (1993)) for a 5'-uORF is stronger relative to what is presumably the main ORFs, suggesting that the rate of translation may be very low. Moreover, the initiation codon of *GhMYB5*'s 5'uORF is separated from the main start codon by only a single nucleotide, which generates two overlapping ORFs. The physical proximity of AUGs may conceivably generate competition between the two ORFs for ribosomal machinery, thus affecting the rate of GhMYB5 synthesis. The physical proximity of AUGs may conceivably generate competition between the two ORFs for ribosomal machinery, thus affecting the rate of GhMYB5 synthesis.--

B15

Please replace the paragraph beginning at page 32, line 13, with the following:

--Based on multiple sequence alignment algorithms, the DBD amino acid sequences of cotton MYB proteins show structural similarity to plant R2R3-MYB factors implicated in phenylpropanoid biosynthesis. Based on the extensive phylogenetic analysis performed by Romero, L. *et al., Plant J* **14**:273-284 (1998), and which also includes GhMYB1 (formerly GhMYBA), GhMYB proteins invariably belong to group C and therefore likely exhibit a preference for type IIG DNA-binding sites. GhMYB5 is the most distantly related cotton R2R3-MYB and is found in an isolated cluster that includes the drought-inducible AtMYB2 (Urao *et al.,* 1996). Amino acid comparisons of DBD and TRR domains from GhMYBs MIXTA (AmMYBMx) and GI1 (AtMYBGIl) did not reveal any striking similarity beyond conserved motifs. However, based on established DBD structural similarities to other R2R3-MYB factors, GhMYB2, GhMYB3, and GHMYB4 are members of a phylogenetic group that contains Glabrous1, while GhMYB1 and GhMYB6 belong to a closely related cluster (Romero, L. *et al., Plant J* **14**:273-284 (1998)). Considering that both Arabidopsis leaf and cotton seed trichomes are single cells, the phylogenetic relationship between Glabrous1 and GhMYBs is especially intriguing from a functional standpoint.--

B16

Please replace the paragraph beginning at page 32, line 29, with the following:

--The general trend found among plant species is for a large number of *MYB* genes (Romero, L. *et al., Plant J* **14**:273-284 (1998); Lin, Q. *et al., Plant Mol Biol* **30**:1009-1020 (1996); Solano, R. *et al., Plant J* **8**:673-682 (1995b)). Genomic Southern blots hybridized with *GhMYB* DBDs established that this is also the case in cotton. Yet, DNA blots hybridized with gene-specific probes indicate each *GhMYB* class is encoded by a small gene family consisting of only a relatively few number of genes. The DNA blots probably represent the minimum number of genes belonging to each family, since it is a strong possibility that the hybridization pattern does not distinguish alloalleles derived from of the AA and DD genomes of *G. hirsutum* (Wilkins, T.A. *et al., Theor Appl Genet* **89**:514-524 (1994)). However, the increase in ploidy level likely results in a simple amplification of the number of genes in each family. Thus, the six novel *GhMYB* genes identified in this study represent only a small subset of the *MYB* genes encoded by cotton genome.--

B17

Please replace the paragraph beginning at page 33, line 8, with the following:

--A semi-quantitative RT-PCR approach (An, Y-Q. *et al., Plant Cell* **8**:5-30 (1996)) proved to be a key means to characterizing the differential expression patterns for each of the six *GhMYB* genes since transcript abundance was too low to be detected by conventional RNA blot analysis. RT-PCR experiments revealed that the spatial and temporal regulation of all six *GhMYBs* form two distinct patterns of gene expression. Type I *GhMYB* (*GhMYB1*, *-2* and *-3*) transcripts were more abundant than type II genes and were found in all tissue-types examined, suggesting that type I cotton *MYBs* regulate cellular functions common to all these tissues. In contrast, type II cotton *GhMYBs*, (*GhMYB4 -5,* and *-6*) are not only spatially and temporally modulated to a greater degree than type I *GhMYBs*, these mRNAs are much less abundant than type I messages since type II transcripts are detected only at the lower RT dilutions for the most part. As suggested previously (Jackson, D. *et al., Plant Cell* **3**:115-125 (1991); Larkin, J.C. *et al., Plant Cell* **5**:1739-1748 (1993); Solano, R. *et al., EMBO J* **14**:1773-1784 (1995a)) for greater other plant MYB genes. the spatial and temporal modulation of type II *MYBs* suggests that these MYBs may be involved in modulating the fine-tuned control of specific cellular functions. Interestingly, the distinct pattern of transcript accumulation observed for *GhMYB4* and *GhMYB5* and the presence of putative 5'-uORFs in the 5'-untranslated region suggests that the spatial and temporal expression of these type II cotton *MYBs* are subject to complex regulation at both the transcriptional and post-transcriptional levels.--

Please insert the accompanying paper copy of the Sequence Listing, page numbers 1 to 14, at the end of the application.

REMARKS

Applicants request entry of this amendment in adherence with 37 C.F.R. §§1.821 to 1.825. This amendment is accompanied by a floppy disk containing the above named sequences, SEQ ID NOS:1-26, in computer readable form, and a paper copy of the sequence information which has been printed from the floppy disk.

The information contained in the computer readable disk was prepared through the use of the software program "PatentIn" and is identical to that of the paper copy. This amendment contains no new matter.